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ARAŞTIRMA MAKALESİ **RESEARCH ARTICLE**

Isolation of endophytic bacterial isolates from healthy banana trees and determination of their *in vitro* antagonistic activities against crown rot disease agent *Fusarium verticillioides*

Sağlıklı muz ağaçlarından endofit bakteri izolatlarının izolasyonu ve taç çürüklüğü hastalığı etmeni *Fusarium verticillioides*'e karşı antagonistik etkinliklerinin *in vitro* koşullarda belirlenmesi

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Ö Z E T / A B S T R A C T

Aims: Fungal diseases are one of the most important biotic factors causing serious losses in banana cultivation in field or greenhouses during cultivation, harvest, storage and transportation periods. Crown rot, caused by *Fusarium verticillioides,* is considered one of the most important postharvest fungal disease of banana fruits. In this study, endophytic bacterial isolates were obtained from fruits, branches and leaves of healthy banana trees and their antagonistic potentials were investigated against *F. verticillioides* as a biological control agent (BCA) *in vitro* conditions.

Methods and Results: A total of 23 putative endophytic bacterial isolates were obtained from fruits, stems and leaves of healthy banana trees. All bacterial isolates were identified by MALDI-TOF MS (Matrix Assisted Laser Desorption Ionization-Time Of Flight Mass Spectrometry) analysis. *In vitro* antagonistic efficiencies of 12 different bacterial isolates, from eight different species (*Bacillus subtilis* ssp. *spizizenii* (3), *Bacillus amyloliquefaciens* (2), *Bacillus subtilis* ssp. *subtilis* (2), *Bacillus mojavensis*, *Enterococcus faecium, Enterobacter cloacae, Enterobacter ludwigii* and *Pseudomonas stutzeri*), were tested on inhibiting mycelial growth by using dual culture tests. Among bacterial isolates, the highest antagonistic activity was displayed by *Bacillus mojavensis* BEn3 isolate which significantly inhibited the mycelial growth by 50.83%. *Enterobacter cloacae* BEn1, *Enterobacter ludwigii* BEn2, *Enterobacter faecium* BEn7, however, failed to inhibit the mycelial growth of fungi.

Conclusions: Significant suppression in the mycelial growth caused by endophytic bacterial isolates indicates that *Bacillus mojavensis* BEn3 isolate could be considered as possible BCA against crown rot disease agent.

Significance and Impact of the Study: Our findings suggest that *B. mojavensis* Ben3, as the most successful endophyte bacterial isolate that suppresses the growth of *F. verticillioides*, can be used as a promising biological control agent as an alternative to chemical control against crown rot disease.

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INTRODUCTION

Although the banana is a tropical climate fruit, it is also grown in subtropical climatic conditions in some microclimates. In Turkey, banana production meets almost 50% of domestic consumption amount. The difference between consumption and production is met through imports and five international companies control 80% of the world's banana trade. While the average annual yield of Turkey is 6.528 kg/da of bananas, the production amount was 728.133 tons in 2020. According to TUIK data, 1.101 da of this area are located in the provinces of Hatay Province (Anonymous, 2021). In recent years, there has been a rapid transition to greenhouse cultivation in the mentioned areas and a significant increase in production and quality has been achieved. The production of bananas is the main source of livelihood for 45% of the people living in the areas where the banana grows (Pınar et al., 2011).

As a climacteric fruit, banana have high nourishment and are rich in nutrient contents such as other ripe fruits, very short shelf life. Mature fruits are susceptible to many postharvest disease agents which cause high rates of loss (Maqbool et al., 2011). Fungal disease agents are the most important biotic factors causing serious loss during cultivation, storage and transportation. Nowadays, postharvest fruit loss caused by fungal disease agents is estimated to account for more than 50% of total agricultural fruit production (Zhang et al., 2017). Crown rot, anthracnose, Lasiodiplodia rot, Thielaviopsis rot, cigar-end rot, black-tip rot and squirter were reported to be the most common diseases in the world's leading banana growing countries (Snowdon, 2010).

In banana fruits, decaying of the region called the crown (chock) is known as crown rot disease. Crown rot disease complex was reported to be caused by various fungal species either alone or more than single species. Following morphological and molecular identification studies, Fusarium species were the most frequently isolated and reported fungal agents responsible for crown rot disease. Since disease agents cause considerable economical losses during transportation and storage stages, researchers in banana producing countries have begun to give priority to effective control methods against the disease agents observed in the banana fruit (Ranasinghe et al., 2002; Anthony et al., 2004; Williamson et al., 2008; Umana-Rojas and Garcia, 2011; Alvindia, 2013; Appiah-Sarpong et al., 2013). In a very recent study, several fungal disease agents were isolated and identified on banana fruits in storage and packaging houses located in Mersin province of Turkey. Among the fungal isolates, obtained from the fruits showing crown rot disease symptoms, isolates belonging to Fusarium spp. such as *Fusarium verticillioides* (13.4%), *F. oxysporum* (10.5%), *F. proliferatum* (9.7%) were the most frequently isolated fungal agents responsible for crown rot disease (Faruk and Soylu, 2021). Fungicides are the most commonly used control strategy for controlling banana leaf spots and post-harvest diseases. Triazoles and prochloraz were reported as highly effective fungicides for suppressing these diseases. Overuses of fungicides were, however, frequently resulted in chemical residues in banana plants and their environments which become a public concern. Therefore, eco-friendly methods are necessary for reducing fungicide usage in the management of banana diseases (Fu et al., 2010).

Biological control, using bacterial microbiomes, was frequently reported great promise of providing an alternative to current postharvest chemical fungicides for the control of postharvest diseases and reducing the health and environmental hazards presented by synthetic fungicides (Wilson et al., 2011; Dukare et al., 2019; Ons et al., 2020; Tariq et al., 2020).

Bacterial microbiomes, used as biological control agents (BCA) and plant growth promoting (PGPB), are named as endophytes or epiphytes according to their location in the plant. Both BCA and PGPB isolates have been used to prevent several kinds of plant diseases in the context of biological control. Endophytic bacteria are defined as bacteria isolated from the internal tissues of surface disinfected plant parts such as flower, leaf, fruit, stem, root and seed and not causing any damage to the treated plants (Zhang et al., 2004; Anand et al., 2010; Akram and Anjum, 2011; Tan et al., 2012; Eljounaidi et al., 2016; Santoyo et al., 2016; Sülü et al., 2016). Bacterial isolates belonging to *Agrobacterium, Bacillus, Burkholderia, Chryseobacterium, Clavibacter, Curtobacterium, Enterobacter, Micrococcus, Paenibacillus, Phyllobacterium, Rhizobium, Pseudomonas, Serratia and Stenotrophomonas genera* are known as endophytic antagonistic bacteria (McInroy and Kloepper, 1995).

Endophytic antagonistic bacteria have been reported to possess several disease suppressive mechanisms, such as antibiosis, siderophore production, carbon source competition and plant resistance, to prevent pathogenesis (Eljounaidi et al., 2016). Endophytic bacterial isolates have been also reported promoting plant growth by activating the expression of several hormones in the plant, resulting in healthy development of the plant, resistance to disease agent(s) and an increase in the yield (Santoyo et al., 2016). The use of bacterial microbiomes as biocontrol agents (BCA) has

been very important in the integrated management of cultivations and organic production, where their value as a postharvest control of fungal diseases stands out. Bacterial isolates of *Burkholderia cepacia* (De Costa and Erabadupitiya, 2005), *Bacillus amyloliquefaciens* (Alvindia and Natsuaki, 2009), *Pseudomonas syringae* (Williamson et al., 2008), *Pantoea agglomerans* and *Flavobacterium sp*. (Gunasinghe and Karunaratne, 2009) were reported for controlling banana post-harvest diseases in previously conducted biocontrol studies. The roles of endophytes in disease and pest resistance are comparatively understudied, but recent work has started to highlight the importance of endophytes, in particular, as an increasingly popular biological control option (Gao et al., 2010; Dutta et al., 2014; Bozkurt and Soylu, 2019; Rabiey et al., 2019).

The multi purposes of this study were to (i) isolate endophytic bacterial isolates, having biocontrol potential from the healthy banana internal tissues of the fruits, stems and leaves, to (ii) identify into species level, and to (iii) investigate their antagonistic efficiencies against crown rot disease agent F*. verticillioides in vitro* conditions.

MATERIALS and METHODS

Isolation of fungal disease agent

The fungal disease agent *F. verticillioides* was originally isolated from the banana fruits (cv. Azman) displaying typical crown rot symptoms (Figure 1) and purified on the PDA medium (Kurt et al., 2020a,b; Faruk and Soylu, 2021). Carnation Leaf-Piece Agar (CLA) medium was used for identification of fungal species with the reason of forming uniform micro- and macroconidia, chlamydospore production as well as colony morphology, pigmentation and growth rates on the medium (Fisher et al., 1982; Leslie and Summerell, 2006).

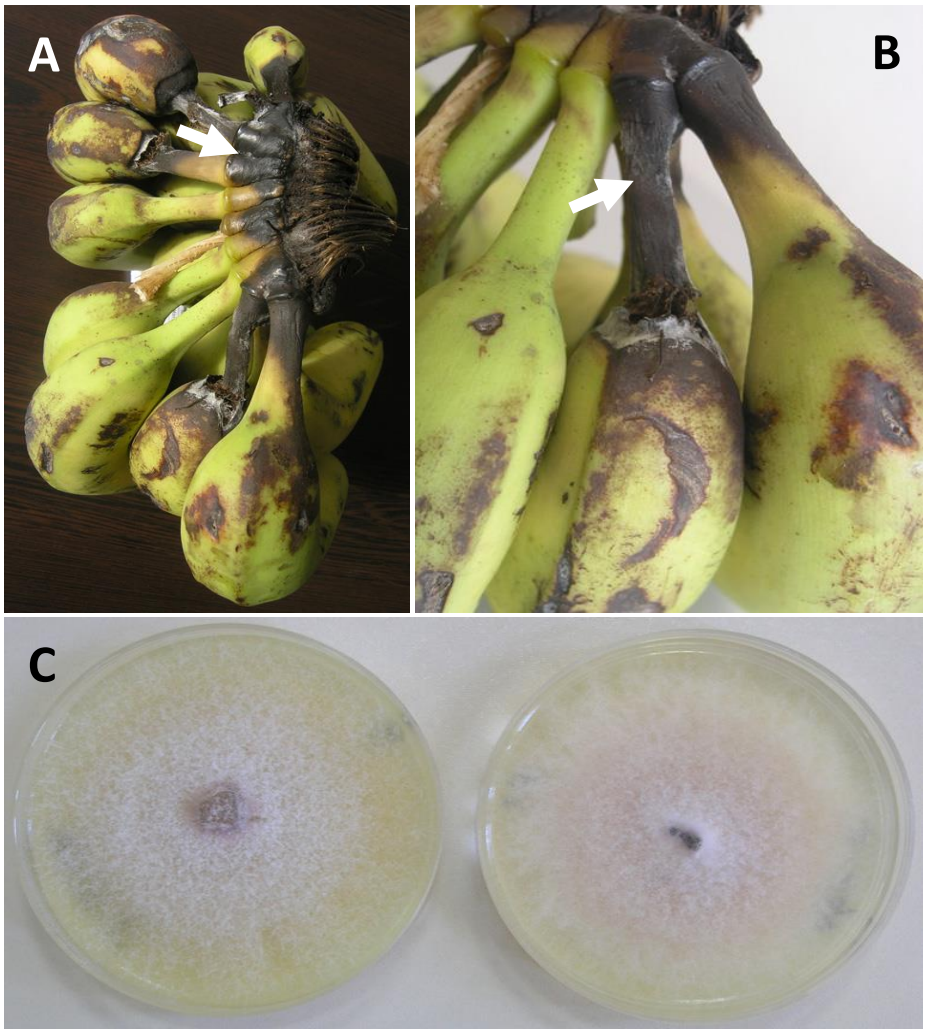

Figure 1. (A and B) Typical softening and sporulation symptoms (arrows) caused by crown rot disease agent *F. verticillioides* on banana fruits. (C) Typical mycelial growth of *F. verticillioides* on PDA nutrient medium

Petri dishes were then incubated for 7-10 days at 25°C and the identification of disease agent was made according to morphological characteristics such as colony growth and pigmentation, spore shape and color, chlamydospore formation, conidiophore structure as described by Leslie and Summerell (2006) under light microscopy (Olympus BX 51). Identification of fungal isolate was further confirmed by MALDI-TOF MS protein profile analyses as described previously (Kurt et al., 2017; Soylu et al., 2021).

Isolation of endophytic biocontrol bacterial isolates

Endophytic bacteria were obtained from internal tissues of surface sterilized fruits, stems and leaves of healthy banana trees growing in Arsuz district of Hatay province (Figure 2). Surface sterilized fruits and stems were cut with sterile bisturi and isolation was made on the nutrient medium through directly touching crushed internal tissues (imprinting method). The surface-sterilized leaves were crushed directly in sterile phosphate buffered saline solution (pH 7.4) and the resulting suspension was transferred onto selective King B Agar (KB) or Tryptic Soybean Agar (TSA) medium (Aktan ve Soylu, 2020). The cultured petri dishes were then kept at 26°C for 48 hours. Pure bacterial isolates were obtained from bacterial colonies with different morphological appearances growing on the surface of the medium. Each of the bacterial colonies with different morphological appearances representing the petri dishes was evaluated as a putative antagonist bacterial isolate (Kara ve ark., 2020). To determine whether the endophytic bacterial isolates are plant pathogen, all putative isolates were subjected to the hypersensitivity (HR) test in the tobacco plant (Schaad, 2001). Isolates that do not produce HR are considered as putative endophytic bacterial isolates and maintained in petri dishes containing TSA medium at +4°C until diagnosis.

Figure 2. General appearances of healthy banana fruit trees from which endophytic bacterial isolates were obtained. Bacterial isolates were especially obtained from healthy fruits (*) adjacent to diseased fruits (arrow)

Identification of endophytic biocontrol bacterial isolates by MALDI-TOF MS analyses

The identification of the candidate endophytic bacterial isolates was first made according to the biochemical methods (Lelliot and Stead, 1987; Schaad, 2001). Bacterial isolates were then identified precisely into species by using Matrix Assisted Laser Desorption İonization-Time of Flight Mass Spectrometry (MALDI-TOF MS) analyses (Aktan ve Soylu, 2020). Protein isolation was made from putative antagonist endophytic bacterial isolates which were derived from purified from single colonies by using the ethanol-formic acid extraction method. The specific protein spectra of the isolates were obtained with Maldi Biotyper Real-Time Classification (RTC) software (Biotyper 3.0; Microflex LT; Bruker Daltonics GmbH, Bremen, Germany). Bacterial

isolates were then identified at species levels with a high-confidence score by comparing them with the spectra of the reference culture species in the library. At the end of the analysis, a score value of 2.30-3.00 (green color) was considered as the highly probable species identification, score value of 2.00-2.299 (green color) was considered as secure genus identification and probable species identification, score value of 1.70-1.99 (yellow color) was considered as probable genus identification and a score of 0.00-1.69 (red color) were considered as an unreliable diagnosis.

Determination of activities of antagonist endophytic bacteria in vitro conditions

Antagonistic potentials of endophytic bacteria obtained from the internal tissues of fruits, branches and leaves were determined by dual culture tests in PDA-containing petri dishes in vitro conditions as previously reported (Soylu ve ark., 2020). In dual culture tests, the putative endophytic bacterial isolate to be tested was drawn on one end of the media and left for pre-incubation at 26°C for 2 days. After the bacteria developed, mycelial discs with a diameter of 5 mm, taken from the actively growing tip of the 5-day-old fungus culture, were placed 4 cm away from the developing colony and the petri plates were left to develop at 26°C. As a control, the fungus was transferred to non-bacterial petri dishes. As the mycelial growth of the fungus reached the determined point in the control petri dishes (MGc), fungal mycelial growths towards the bacteria were measured in all petri dishes containing putative endophytic antagonist bacterial isolates (MGb) and the % inhibition rates were calculated according to the mycelial growth in the control petri dishes by using %Abbot formula (Inhibition % = [(MGc-MGb) / MGc) * 100] as decribed by Soylu et al. (2021).

In vitro biocontrol experiments were set up according to the randomized plot design, with 3 replications for each application, and repeated at 2 different times. One-way ANOVA analysis was performed on the mycelial inhibition values in the petri dishes using the SPSS statistical program (SPSS Statistics 17.0), and the difference between the isolates were analyzed with the Duncan Multiple Range Test (P≤0.05).

RESULTS and DISCUSSION

Fungal isolate was obtained from surfaces of blackened and covered with whitish mycelia on banana crown pedicels (Figure 1A, B). Colonies grew rapidly with abundant pink to violet aerial hyphae in petri dishes containing PDA medium (Figure 1C). Fungal isolate was identified according to morphological characteristics of the developing colonies, micro and macroconidia, sporodochia, conidiophores, phialides. No chlamydospores observed. Based on morphological characteristics, the disease agent was tentatively identified as *F. verticillioides* (Al-Hatmi et al., 2016). Identification of fungal isolate was further confirmed by MALDI-TOF analysis. Representative fungal isolate was tested for pathogenicity and found to be casual disease agent of crown rot on banana fruit (Faruk and Soylu, 2021) and used for biocontrol studies.

A total of 23 putative bacterial isolates were obtained from the inner tissues of healthy fruits, stems and leaves of banana trees obtained from the different greenhouses located in Arsuz district of Hatay province (Figure 2). These isolates obtained were subjected to several tests to determine whether putative bacterial isolates were plant or human pathogenic. As a result of the test, 18 bacterial isolates out of 23 candidate bacteria did not cause HR at the inoculation point. Since five isolates caused soft rot on potato slices and one isolate grew at 37°C among 18 isolates, and 6 isolates were excluded from the trials due to the possibility of potential plant and human pathogens. The remaining twelve isolates, obtained from healthy tissue, were then identified at species level by MALDI-TOF MS analyses.

MALDI-TOF MS analyses of 12 isolates (coded as BEn1 to BEn12) revealed the species identification (according to the isolate order) as *Enterobacter cloacae, Enterobacter ludwigii, Bacillus mojavensis, Bacillus subtilis ssp. spizizenii, Bacillus subtilis ssp. spizizenii, Pseudomonas stutzeri, Enterococcus faecium, Bacillus amyloliquefaciens, Bacillus amyloliquefaciens, Bacillus subtilis ssp. spizizenii, Bacillus subtilis ssp. subtilis and Bacillus subtilis ssp. subtilis*.

The *in vitro* antagonistic potentials of 12 putative endophytic antagonist bacteria isolates were, then, investigated against the crown rot disease agent *F. verticilloides*. Isolates belonging to different *Bacillus spp*. were inhibited at high rates such as 39.17-50.83% (Table 1, Figure 3). Among antagonist bacterial isolates tested, the most effective suppression of mycelial growth was caused by *B. mojavensis* BEn3 isolate (suppression up to 50.83%). *Pseudomonas stutzeri* (BEn6), another antagonist species other than Bacillus species, also inhibited the mycelial growth at rate of 36.67% which was statistically different from Bacillus species (Table 1). Different isolates belonging to *Enterobacter spp.* such as *Enterobacter cloacae* (BEn1), *Enterobacter ludwigii* (BEn2), *Enterococcus faecium* (BEn7) were, however, failed to inhibit the mycelial growth of fungal disease agent.

Table 1. Inhibition potential of antagonist endophytic bacterial isolates obtained from the internal tissues of banana fruits and leaves to the growth of fungal disease agent *F. verticillioides* (%) *in vitro* conditions[a]

Isolate Nu.	Species name	Mycelial growth (mm)[b]	Inhibition of fungal growth (%)
BEn1	*Enterobacter cloacae*	40.00a	0.00
BEn2	*Enterobacter ludwigii*	40.00a	0.00
BEn3	*Bacillus mojavensis*	19.67e	50.83
BEn4	*Bacillus subtilis* ssp. *spizizenii*	20.33de	49.17
BEn5	*Bacillus subtilis* ssp. *subtilis*	20.67de	48.33
BEn6	*Pseudomonas stutzeri*	25.33bg	36.67
BEn7	*Enterococcus faecium*	40.00a	0.00
BEn8	*Bacillus amyloliquefaciens*	20.67de	48.33
BEn9	*Bacillus amyloliquefaciens*	20.67de	48.33
BEn10	*Bacillus subtilis* ssp. *spizizenii*	21.33d	46.67
BEn11	*Bacillus subtilis* ssp. *subtilis*	24.33bc	39.17
BEn12	*Bacillus subtilis* ssp. *subtilis*	23.00c	42.50
Control	***Fusarium verticillioides***	40.00a	0.00

[a] Bacterial isolates were scratched on the PDA 48 hours before than pathogen inoculation. The mycelial growth (mm) of the fungus towards the bacterial pathway was measured in the petri dishes on which the bacteria were found and compared with the growth on the control petri dishes. Then the inhibition rates (%) were calculated.

[b] The values obtained were averages of measurements in 3 different petri dishes and the experiment was repeated twice at different times. Mean values followed by different letters in the column indicate that the difference between isolates is statistically significant (Duncan Multiple Comparison Test. P <0.05).

Recently, important information and advances concerning the isolation, identification mode of action, different approaches to enhance biocontrol activity, formulation and production of beneficial microorganisms such as antagonistic bacteria have been achieved and commercialised biopesticides are already in the market (Fravel, 2005). Nonetheless, it is necessary to continue finding new potential microbiomes, better understanding the mode of action, and pathogen, antagonist and host interactions, to increase the potential of biocontrol helping to become a real alternative to synthetic postharvest fungicides (Nunes, 2012).

It is known that the Bacillus species, which have several antagonistic properties, are the most emphasized species in biological studies due to their ability to synthesize antibiotic substances (Stein, 2005). It has been reported that *B. subtilis* is the most suitable microbial species for the production of biopesticides due to its biological properties such as producing antimicrobial compounds with different molecular structures, pathogens cannot easily develop resistance against the produced compounds, and forming endospores that enable it to survive under severe environmental conditions (Fravel, 2005; Abbas et al., 2019).

Alvindia and Natsuaki (2009) isolated two bacteria which were identified as *Bacillus amyloliquefaciens*, from the surface of banana fruits. Both isolates were screened for in vitro antagonism toward crown rot disease agents *Lasiodiplodia theobromae, Thielaviopsis paradoxa, Colletotrichum musae* and *Fusarium verticillioides* and were found to be antagonistic.

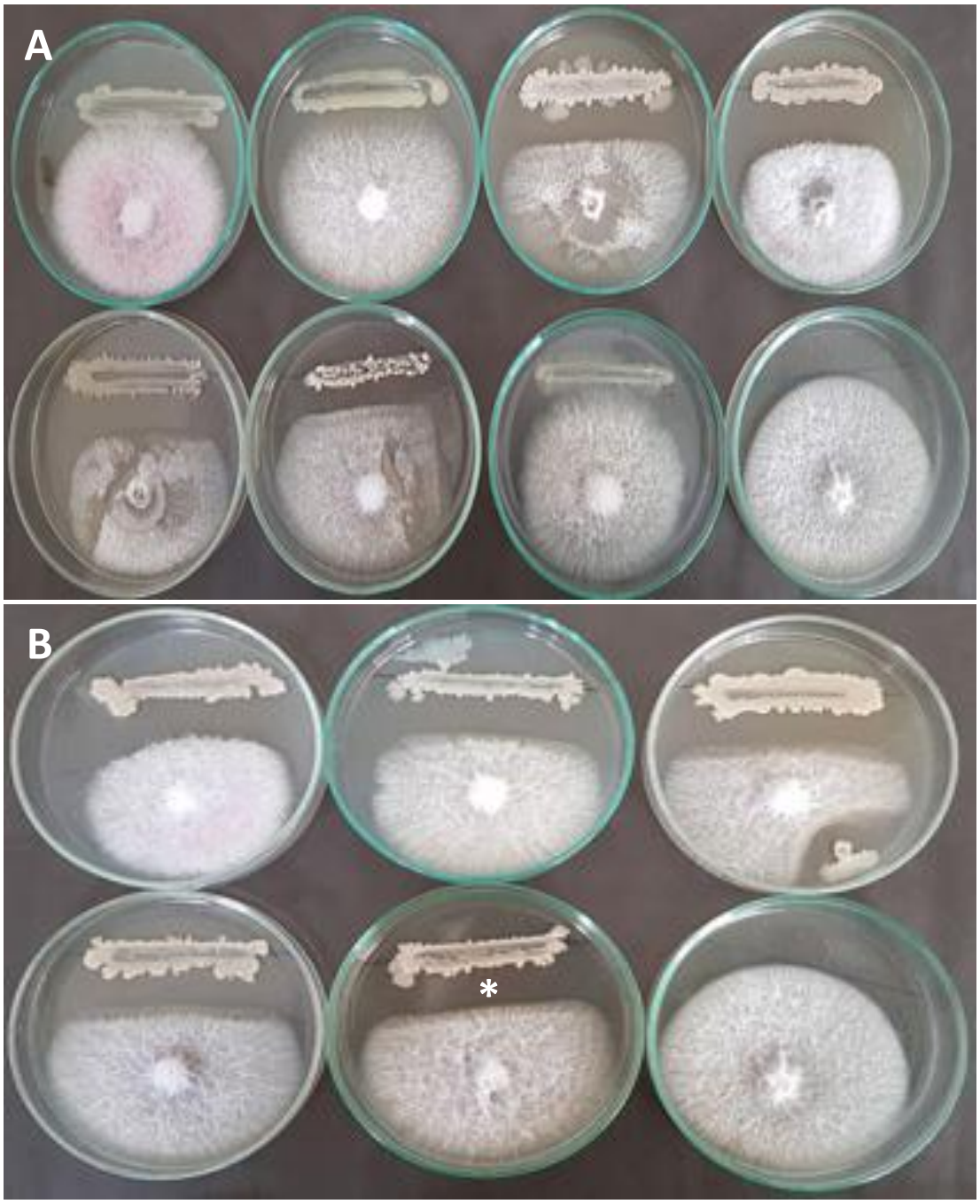

Figure 3. (A) *In vitro* antagonistic (inhibitory) effect of different antagonistic bacterial isolates on mycelial growth of fungal agent *F. verticillioides*. Inefficient isolates (Ben1, Ben2, Ben7) typically failed to inhibit mycelial growth. (B) Shows efficient isolates (Ben3, Ben4, Ben9, Ben11, Ben12) efficiently inhibited mycelial growth causing inhibition zones (*) between bacteria and fungal mycelia

The *B. amyloliquefaciens* DGA14 isolate produced a diffusible metabolite that inhibited all test pathogens in culture. In addition, the bacterial isolate was found to be significantly affecting mycelial growth and conidial germination in a liquid medium. Postharvest application of *B. amyloliquefaciens* DGA14 in the packing house reduced the incidence of crown rot to a level significantly lower than in fungicide treated or control fruits. Fu et al. (2010) also characterized bacterial biocontrol strain B106, identified as *Bacillus subtilis*, from rhizospheric soil of a banana plant and determined its efficacy in controlling banana leaf spot and post-harvest anthracnose diseases. The efficacy of bacterial isolate *Bacillus subtilis* B106 in controlling both banana leaf spot diseases in the field and anthracnose disease at post-harvest stage was determined as 48.3% and 48.6%, respectively. Their results clearly showed that the antagonistic *Bacillus subtilis* strain B106 was a promising

biocontrol agent against banana diseases. In the recent study conducted by Damasceno et al. (2019), biocontrol potentials of 12 putative bacterial isolates, which were selected from *in vitro* tests, were evaluated *in vivo* testing. Among bacterial isolates, *Bacillus velezensis, Enterobacter cloacae, Serratia marcescens* and *Stenotrophomonas maltophilia, B. velezensis* were found to be effective as similar as the fungicide Thiabendazole. *B. velezensis* inhibited mycelial growth and pathogen sporulation and was positive for cellulase and xylanase activity, which are mechanisms of action relevant to a biocontrol agent. Since *B. velezensis* is not pathogenic to humans, this bacterium was reported to be considered as BCA of direct application by spraying and fruit immersion or can be used systemically.

Although bacterial isolates of *Bacillus subtilis, Bacillus amyloliquefaciens* and *Bacillus velezensis* were previously reported to have biocontrol potentials against post-harvest fungal disease agents of banana such as *Lasiodiplodia theobromae, Colletotrichum musae, Thielaviopsis paradoxa* and *Fusarium verticillioides* (Alvindia and Natsuaki, 2009; Sangeetha et al., 2010; Fu et al., 2010; Damasceno et al., 2019), this is the first report of *B. mojavensis* as a biocontrol agent against the banana crown rot disease agent caused by *Fusarium verticilloides*. In the recent study conducted by Atay et al. (2020), antifungal activities of 12 putative endophytic bacterial isolates (*Bacillus pumilus, Bacillus subtilis ssp. subtilis, Bacillus amyloliquefaciens, Bacillus vallismortis, Bacillus mojavensis, Bacillus megaterium, Solibacillus silvestris, Erwinia herbicola, Corynebacterium glutamicum, Bacillus cereus, Pantoea dispersa* and *Bacillus endophyticus*) were tested against postharvest heart rot disease agent *Alternaria alternata* on pomegranate fruits by using in dual culture assays. Among the tested isolates, *Bacillus mojavensis* PEB39 was the most effective isolate against pathogenic fungi *in vitro* (80% inhibition of mycelial growth) followed by *Bacillus amyloliquefaciens* PEB46 (78.9%) *Bacillus vallismortis* PEB40 (76.7%) and *Bacillus subtilis* ssp. *subtilis* PEB43 (75.6%), respectively (Atay et al., 2020).

In our study, although the mechanism(s) involved in mycelial growth inhibition was not studied, the existence of an inhibition zone surrounding bacterial isolates indicated the production of unknown antibiotic substance(s) by the antagonist isolates.

In conclusion, endophytic antagonist microorganisms isolated from the internal tissues of healthy plants are known to have a higher chance of success in biological control as they require faster adaptation or minimal growth conditions compared to microorganisms developed in laboratory conditions. According to this study, the endophyte antagonistic bacterial isolates isolated from healthy plant tissues are effective on the disease conditions after harvest and will increase the quality and income of crops by decreasing the pesticide application which is used intensively and frequently against the plant diseases after harvest. Using these environmentally friendly, biologically based microorganisms, the reduction of pesticide applications in the production areas will greatly contribute to the adverse effects of pesticides on the environment and human health.

ÖZET

Amaç: Fungal hastalıklar tarla veya seralarda muz yetiştiriciliğinde ekim, hasat, depolama ve nakliye dönemlerinde ciddi kayıplara neden olan en önemli biyotik faktörlerden biridir. *Fusarium verticillioides*'in neden olduğu taç çürüklüğü, muz meyvelerinin en önemli hasat sonrası fungal hastalıklarından biri olarak kabul edilir. Bu çalışmada, sağlıklı muz ağaçlarının meyve, dal ve yapraklarından endofitik bakteri izolatları elde edilmiş ve biyolojik kontrol ajanı (BCA) olarak antagonistik potansiyelleri *F. verticillioides*'e karşı in vitro koşullarda araştırılmıştır.

Yöntem ve Bulgular: Sağlıklı muz ağaçlarının meyvelerinden, gövdelerinden ve yapraklarından toplam 23 adet aday endofit bakteri izolatı elde edilmiştir. Tüm bakteri izolatları MALDI-TOF MS (Matrix Assisted Laser Desorpsiyon Ionization-Time Of Flight Kütle Spektrometresi) analizi ile tanımlanmıştır. Sekiz farklı türden 12 farklı bakteri izolatının (*Bacillus subtilis* ssp. *spizizenii* (3), *Bacillus amyloliquefaciens* (2), *Bacillus subtilis* ssp. *subtilis* (2), *Bacillus mojavensis*, *Enterococcus faecium*, *Enterobacter cloacae*, *Enterobacter ludwigii* ve *Pseudomonas stutzeri*) misel büyümesinin engellenmesindeki *in vitro* antagonistik etkinlikleri ikili kültür testleri ile test edilmiştir. Bakteri izolatları arasında en yüksek antagonistik aktivite misel gelişimini %50.83 oranında engelleyen *Bacillus mojavensis* BEn3 izolatı tarafından gösterilmiştir. Bakteriyel izolatlar arasından *Enterobacter cloacae* BEn1, *Enterobacter ludwigii* BEn2, *Enterobacter faecium* BEn7 izolatları fungal etmenin misel gelişimini engellemede başarısız olmuştur.

Genel Yorum: Endofit bakteri izolatlarının fungal etmenin misel gelişimini önemli düzeylerde baskılamış olması, *Bacillus mojavensis* BEN3 izolatının taç çürüklüğü etmenine karşı olası biyolojik kontrol ajanı olarak kabul edilebileceğini göstermektedir.

Çalışmanın Önemi ve Etkisi: Bulgularımız, *F. verticillioides*'in gelişimini baskılayan en başarılı endofit

bakteri izolatı olan *B. mojavensis* Ben3'in taç çürüklüğü hastalığına karşı kimyasal mücadeleye alternatif ve umut verici bir biyolojik kontrol ajanı olarak kullanılabileceğini önermektedir.

Anahtar Kelimeler: Muz, Biyolojik mücadele, endofit bakteri, taç çürüklüğü, *Fusarium verticillioides*.

CONFLICT OF INTEREST

The authors declare no conflict of interest for this study.

AUTHOR'S CONTRIBUTIONS

The contribution of the authors is equal.

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